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Anti-bacterial studies on *Hemigraphis colorata* (Blume) H.G. Hallier and *Elephantopus scaber* L.

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ABSTRACT

Objective: To examine the ethanol, aqueous, chloroform, benzene, acetone and petroleum ether extracts of *Hemigraphis colorata* (*H. colorata*) leaves and stem and *Elephantopus scaber (E. scaber)* leaves, root and flower for the presence of phyto-constituents and screened the anti-bacterial activity against the selected pathogens. **Methods:** The fresh materials were shade dried and powdered using the tissue blender. The dried and powered materials (50 g) were extracted successively with 200 mL of aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether by using Soxhlet extractor for 8 h at a temperature not exceeding the boiling point of the solvent. Aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts were prepared from powdered materials were used for preliminary phytochemical and antimicrobial studies using standard methods. **Results:** The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root and *H. colorata* leaves and stem demonstrated that out of (5×6×12 = 360) tests for the presence or absence of the above compounds, 188 tests gave positive results and the remaining 172 gave negative results. The results of the phytochemical screening revealed that phenol (12/12), carbohydrates (9/12), steroids (8/12), saponins and coumarins (7/12), tannins (6 /12), proteins (5/12), carboxylic acid and flavonoids (4/12), xanthoproteins (3/12) and alkaloids (2/12) presence in the crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *H. colorata* leaves and stem. The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root displayed the presence of phenol (18/18), tannin (17/18), carbohydrates (16/18), steroids (14/18), carboxylic acid and coumarins (12/18), saponins (10/18), xanthoprotein (9/18), flavonoids (7/18), protein (4/18) and alkaloids (2/18). The root ethanolic extracts of *E. scaber* illustrated the highest zone of inhibition against three pathogens viz., *Staphylococcus aureus* (*S. aureus*) (24 mm), *Escherichia coli (E. coli)* (16 mm) and *Pseudomonas aeruginosa* (*P. aeruginosa*) (13 mm). The chlorofrom extracts of *E. scaber* showed the highest zone of inhibition against *Bacillus cereus* (*B. cereus*) (12 mm), The leaves ethanolic extracts of *E. scaber* demonstrated the highest zone of inhibition against three pathogens viz., *Enterococcus faecalis (E. faecalis)* (18 mm), *Proteus mirabilis* (*P. mirabilis*) (17 mm), *Salmonella Typhi* (*S. typhi*) (14 mm) and *Enterobacter* sp. (11 mm) While the benzene extracts of *H. colorata* demonstrated maximum zone of inhibition against the pathogen *Acinetobacter* sp. (14 mm) and *S. aureus* (12 mm). **Conclusions:** It is hoped that this study would direct to the establishment of some compounds that could be used to invent new and more potent antimicrobial drugs of natural origin.

1. Introduction

The therapeutic efficacies of many indigenous plants, for various diseases have been described by traditional herbal medicine practitioners. Natural products are a source of synthetic and traditional herbal medicine. They are still the primary health care system in some parts of the world. The past decade has been seen considerable change in opinion regarding ethno-pharmacological therapeutic applications. The presence of various life sustaining constituents in plants has urged scientists to examine these plants with a view to determine potential medicinal properties. *Hemigraphis colorata* (*H. colorata*) (Blume) H.G. Hallier. leaf paste when applied on the wound promoted wound healing in mice but oral administration was ineffective. The wound contraction and epithelialisation was faster in the leaf paste applied on mice. Saravanan *et al*[1] studied the wound healing activity of *H. colorata* and found that the better wound healing in test group may be due to

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the increase in collagen concentration per unit area and stabilisation of fibres. The Increased wound con-traction and tensile strength may be due to the active constituents present in the extract and support the use of *H. colorata* in the topical management of wound healing. *H. colorata* is claimed in folk medicine that the plant has very good wound healing activity[2]. The leaves are ground into a paste and applied on fresh cut wounds. In Chinese folk medicine *Elephantopus scaber* (*E. scaber*) L. is widely used in the treatment of nephritis, edema, dampness, pain in the chest, fever and cough of pneumonia, scabies, and arthralgia due to wounding. It is also commonly used as a remedy for the treatment of gastropathy, hepatitis, nephritis, scabies, arthralgia, leukemia, edema, chest pain, fever and cough of pneumonia, bronchitis, arthritis, carbuncle and eliminates bladder stones in China. *E. scaber* roots and leaves are used as emollient for dysuria, diarrhoea, dysentery, swellings and stomach pain. Root is prescribed to prevent vomiting. Powdered with pepper it is applied for tooth-ache. Leaves are used in applications for eczema and ulcers. A number of phytochemicals have also been isolated from this plant, such as ethyl hexadecanoate, ethyl-9, 12-octadecadienoate, ethyl-(Z)-9-octadecenoate, ethyl octadecanoate, lupeol, stigmasterol, stigmasterol glucoside, deoxyelephantopin, 11, 13, dihydrodeoxyelephantopin, lupeol, epifriedelinol, stigmasterol and two new germacranolide sesquiterpene lactones named 17, 19-dihydrodeoxyelephantopin (2) and iso-17, 19-dihydrodeoxyelephantopin[3]. Most of the major studies only involved the bioactivities of the compounds especially deoxyelephantopin[3-9]. In recent years the popularity of complementary medicine has increased. Dietary measures and traditional plant therapies as prescribed by ayurvedic and other indigenous systems of medicine are used commonly in India. The World Health Organization has also recommended the evaluation of the plants effective in conditions where safe modern drugs are lacking[10]. It was further added that the use of plant extracts and phytochemicals with antimicrobial properties may be of importance in therapeutic treatments, whereas in the past few years, a number of studies have been conducted in different countries to prove such efficiencies[11-14]. Different parts of *E. scaber* have been reported to exhibit ethnomedicinal, evaluation of the plant parts revealed the presence of metabolites and antibacterial activity in varied quantities[3-9]. Silja *et al*[2] and Saravanan *et al*[1] reported the ethnomedicinal importance and wound healing property of *H. colorata*. But there is no report on the phytochemical properties and antibacterial activity of *H. colorata* from Western Ghats of Kerala, India. For *E. sacber*, two reports are available on the antibacterial activity against UTI pathogens and multi resistant *Staphylococus aureus* (*S. aureus*). To supplement the previous observation, in the present investigation we examined the ethanol, aqueous, chloroform, benzene, acetone and petroleum ether extracts of *H. colorata* leaves and stem and *E. scaber* leaves, root and flower for the presence of phyto-constituents and screened the anti-bacterial activity against the selected pathogens viz., *Escherichia coli* (*E. coli*), *Klebsiella pneumonia* (*K. pneumonia*), *Pseudomonas aeruginosa* (*P. aeruginosa*), *Staphylococcus aureus* (*S. aureus*), *Bacillus cereus* (*B. cereus*), *Salmonella typhi* (*S. typhi*), *Serratia marcescens* (*S. marcescens*), *Acinetobacter* sp., *Enterobacter* sp., *Proteus mirabilis* (*P. mirabilis*), *Enterococcus faecalis* (*E. faecalis*) and *Streptococcus pyogenes* (*S. pyogenes*).

2. Materials and methods

Healthy, disease free, plant parts *i.e.*, leaves and stems of *H. colorata* (Blume) H.G. Hallier. Leaves, flower and root *E. scaber* L. were collected from Trivandrum District, Kerala, India and authenticated by Dr. P. David Samuel, following identification a voucher specimen of the plant was deposited in the herbarium of Department of Botany, Nesamony Memorial Christian College, Marthandam - 629 165, Tamil Nadu, India. The fresh materials were shade dried and powdered using the tissue blender. The dried and powered materials (50 g) were extracted successively with 200 mL of aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether by using Soxhlet extractor for 8 h at a temperature not exceeding the boiling point of the solvent. The flowers of *E. scaber* were collected from the natural habitat and examined carefully and old, infected, and fungus damaged flowers were removed. 50 g of fresh flowers petals of *E. scaber* was collected and kept in closed conical flask with 200 mL of aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether in a shaker at room temperature for 24 h. After incubation, the extracts were filtered through Whatman No. 41 filter paper and the extracts were collected and stored in the refrigerator at 4 ℃. The aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts were filtered using Whattman filter paper (No.1) and then concentrated in vacuum at 40 ℃ using Rotary evaporator. The residues obtained were stored in a freezer -70 ℃ until further tests[15]. Aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts were prepared from powdered materials were used for preliminary phytochemical and antimicrobial studies. The preliminary phytochemical screening was performed by modified method of Harborne[16-20]. Antimicrobial study was carried out by disc diffusion method[21] against the pathogens viz., *E. coli*, *K. pneumonia*, *P. aeruginosa*, *S. aureus*, *B. cereus*, *S. typhi*, *S. marcescens*, *Acinetobacter* sp., *Enterobacter* sp., *P. mirabilis*, *E. faecalis* and *S. pyogenes*. The inhibition zone and anti-bacterial activity against the pathogenic bacteria were recorded. The experiments were repeated in triplicate and the results were documented.

3. Results

The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *H. colorata* leaves and stem and *E. scaber* leaves, flower and root contained a greater proportion by mass of the metabolites as shown in Table 1. The results of the phytochemical screening revealed that phenol (12/12), carbohydrates (9/12), steroids (8/12), saponins and coumarins (7/12), tannins (6 /12), proteins (5/12), carboxylic acid and flavonoids (4/12), xanthoproteins (3/12) and alkaloids (2/12) presence in the crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *H. colorata* leaves and stem. The crude aqueous, acetone,

benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root displayed the presence of phenol (18/18), tannin (17/18), carbohydrates (16/18), steroids (14/18), carboxylic acid and coumarins (12/18), saponins (10/18), xanthoprotein (9/18), flavonoids (7/18), protein (4/18) and alkaloids (2/18). The quinnone was failed to demonstrate in the aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root and *H. colorata* leaves and stem.

The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root and *H. colorata* leaves and stem demonstrated that out of (5×6×12 = 360) tests for the presence or absence of the above compounds, 188 tests gave positive results and the remaining 172 gave negative results. The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts *E. scaber* leaves, flower and root displayed that out of (3×6×12 = 216) tests for the presence or absence of the above compounds, 121 tests gave positive results and the remaining 95 gave negative results. The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *H. colorata* leaves and stem showed that out of (2×6×12 = 144) tests for the presence or absence of the above compounds, 67 tests gave positive results and the remaining 77 gave negative results.

The crude aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *E. scaber* leaves and root and *H. colorata* leaves and stem were examined for the antibacterial activity against the selected pathogens. The antibacterial activity has been observed in the aqueous, acetone, benzene, chloroform, ethanol, and petroleum ether extracts of *E. scaber* leaves and root and *H. colorata* leaves and stem against all the tested bacteria with varied activity. The maximum zone of inhibition 24 mm for *S. aureus*, 18 mm for *E. faecalis*, 17 mm for *P. mirabilis*, 16 mm for *E. coli*, 15 mm for *Acinetobacter* sp., 14 mm for *S. typhi*, 13 mm for *P. aeruginosa*, 12 mm for *B.cereus*, 8 mm for *S. pyogenes* and *S. marcescens*, 6 mm for *K. pneumoniae*, 20 mm for *P. vulgaris*, 16 mm for *P. aeruginosa* were observed. The root ethanolic extracts of *E. scaber* illustrated the highest zone of inhibition against three pathogens viz., *S. aureus* (24 mm), *E. coli* (16 mm) and *P. aeruginosa* (13 mm). The chlorofrom extracts of *E. scaber* showed the highest zone of inhibition against *B. cereus* (12 mm), The leaves ethanolic extracts of *E. scaber* demonstrated the highest zone of inhibition against three pathogens viz., *E. faecalis* (18 mm), *P. mirabilis* (17 mm), *S. typhi* (14 mm) and *Enterobacter* sp. (11 mm) While the benzene extracts of *H. colorata* demonstrated maximum zone of inhibition against the pathogen *Acinetobacter* sp. (14 mm) and *S. aureus* (12 mm).

4. Discussion

Prusti *et al*[6] used the ethanolic extracts as a solvent for the extraction, Jasmine *et al*[4] used the acetone as a solvent source, Mohan *et al*[9] used chloroform, benzene and methanol as a solvent source. In the present study we used the aqueous, acetone, benzene, chloroform, ethanol and petroleum ether as solvent source for the extraction of the metabolites. Of which the ethanol extracted solvents showed high degree (11/12 pathogens) of antibacterial activity against the selected pathogens. Since the polarity of ethanol is higher, most of the secondary metabolites of *E. scaber* leaves, flower and root and *H. colorata* leave and stem dissolved in ethanol. Flavonoids have been referred to as nature's biological response modifiers because of strong experimental evidence of their inherent ability to modify the body's reaction to allergen, virus and carcinogens. They show anti-allergic, anti-inflammatory, anti-microbial and anti-cancer activity[22, 23]. In the present study we observed the flavonoids presence in *E. scaber* flower chloroform, aqueous, ethanol, acetone, benzene and petroleum ether extracts and *H. colorata* leaves of crude aqueous, chloroform, ethanol and stem of petroleum ether extracts. Tannins are known to possess general antimicrobial and antioxidant activities[24]. Recent reports show that tannins may have potential value as cytotoxic and antineoplastic agents[25]. In the present study we observed the occurrence of the tannin in the crude extracts of both plants. Other compounds like saponins also have anti-fungal properties[26]. Saponins are a mild detergent used in intracellular histochemistry

Table 1
Preliminary phytochemical studies on acetone, benzene and petroleum ether extracts of *H. colorata* and *E. scaber*.

Compounds	Acetone					Benzene					Petroleum ether				
	H. colorata		*E. scaber*			*H. colorata*		*E. scaber*			*H. colorata*		*E. scaber*		
	L	S	L	R	F	L	S	L	R	F	L	S	L	R	F
Alkaloids	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Phenols	3+	3+	3+	3+	3+	3+	3+	3+	3+	3+	2+	3+	3+	2+	3+
Flavanoids	–	–	–	++	+	–	–	–	–	2+	–	+	–	–	+
Saponins	–	–	–	–	–	3+	3+	3+	3+	3+	–	2+	+	–	2+
Proteins	–	–	–	+	–	–	–	–	–	–	–	–	–	–	–
Quinones	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Steroids	–	2+	3+	3+	2+	3+	+	2+	+	+	3+	–	2+	2+	–
Tannins	+	2+	2+	3+	2+	3+	–	2+	3+	2+	3+	–	3+	3+	2+
Xarthoproteins	–	–	2+	3+	–	–	–	–	–	–	–	+	–	–	–
Carboxylic acid	+	2+	2+	–	2+	–	–	3+	–	2+	–	–	–	+	–
Coumarins	2+	2+	2+	2+	2+	–	–	3+	–	2+	–	–	2+	–	–
Carbohydrates	2+	2+	2+	2+	2+	2+	+	+	2+	+	–	–	–	2+	2+
Total	5	6	7	8	7	5	4	7	4	8	3	4	5	5	5

Table 2
Preliminary phytochemical studies on chloroform, aqueous and ethanol extracts of *H. colorata* and *E. scaber*.

Compounds	Chloroform					Aqueous					Ethanol				
	H. colorata		*E. scaber*			*H. colorata*		*E. scaber*			*H. colorata*		*E. scaber*		
	L	S	L	R	F	L	S	L	R	F	L	S	L	R	F
Alkaloids	+	–	–	+	–	3+	–	–	2+	–	–	–	–	–	–
Phenols	3+	3+	3+	2+	3+	2+	3+	3+	3+	3+	3+	3+	3+	3+	3+
Flavanoids	+	–	–	–	+	+	–	–	–	3+	+	–	–	–	2+
Saponins	+	+	–	3+	2+	–	–	–	–	3+	+	+	–	2+	2+
Proteins	2+	+	–	2+	–	–	+	–	–	2+	2+	+	–	++	–
Quinones	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Steroids	+	3+	2+	2+	–	3+	+	2+	–	2+	3+	–	–	3+	+
Tannins	–	–	2+	2+	2+	–	–	2+	+	2+	2+	+	–	3+	2+
Xarthoproteins	–	–	2+	–	–	2+	–	3+	3+	2+	–	3+	+	3+	2+
Carboxylic acid	2+	–	2+	+	+	–	–	–	2+	–	+	–	2+	–	2+
Coumarins	+	+	2+	+	+	–	2+	+	–	2+	+	2+	–	2+	–
Carbohydrates	2+	3+	3+	3+	3+	2+	–	2+	2+	2+	+	+	2+	2+	–
Total	9	6	7	9	7	6	4	6	6	9	9	7	4	8	7

Table 3
Antibacterial efficacy of the plant extracts against human pathogens (zone of inhibition in mm).

Microorganisms	*H. colorata leaf*						*H. colorata stem*						*E. scaber leaf*						*E. scaber* root						Control
	A	C	B	P	E	W	A	C	B	P	E	W	A	C	B	P	E	W	A	C	B	P	E	W	
E. coli	–	7	–	–	–	–	–	7	–	–	–	–	–	–	8	–	15	–	–	–	–	7	16	4	20
K. pneumoniae	–	–	–	–	6	–	–	–	–	–	6	–	–	–	–	–	–	–	–	–	–	–	–	–	20
P. aeruginosa	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	12	–	–	7	–	7	13	–	22
S. aureus	–	–	–	–	–	–	–	–	–	–	–	–	13	11	–	–	18	–	15	12	–	–	24	3	23
B. cereus	–	–	–	–	9	–	–	8	–	–	–	–	7	9	–	7	–	–	–	12	–	9	–	–	–
S. typhi	–	–	8	–	11	–	–	–	–	–	11	–	–	7	–	–	14	–	7	–	–	–	14	–	16
S. marsecens	–	–	–	–	–	–	7	–	–	–	–	–	7	–	–	7	–	–	–	–	7	8	–	–	26
Acinetobacter sp.	–	–	14	10	–	–	–	11	–	10	–	–	10	7	15	–	–	9	–	–	15	12	–	7	22
Enterobacter sp.	–	–	–	–	6	–	–	–	–	–	7	–	–	–	–	–	11	5	–	–	–	–	8	5	21
P. mirabilis	–	6	–	–	9	7	–	8	–	–	11	9	–	–	–	–	17	–	14	–	–	–	15	–	13
E. faecalis	7	–	–	–	–	–	7	–	–	–	–	–	–	–	–	–	18	–	–	–	–	–	16	7	19
S. pyogenes	–	–	–	–	–	–	–	–	–	–	–	–	–	8	–	–	–	–	–	7	–	7	7	–	22
Total	1	2	2	1	5	1	2	4	0	1	4	1	4	5	2	2	7	2	3	4	2	6	8	5	–

A – Acetone; C – Chloroform; B – Benzene, P – Petroleum ether, E – Ethanol, W – Aqueous.

staining to allow antibody access to intracellular proteins. In medicine, it is used in hyper cholestrolaemia, hyperglycemia, antioxidant, anticancer, anti inflammatory and weight loss, *etc*. It is also known to have anti-fungal properties[27]. Saponins have been implicated as bioactive antibacterial agents of plants[28-29]. Plant steroids are known to be important for their cardiotonic activities, possess insecticidal and anti-microbial properties. They are also used in nutrition, herbal medicine and cosmetics. Plant derived natural products such as flavonoids, terpenoids and steroids etc have received considerable attention in recent years due to their diverse pharmacological properties including antioxidant and antitumor activity. Phenolic phytochemicals have antioxidative, antidiabetic, anticarcinogenic, antimicrobial, antiallergic, antimutagenic and anti-inflammtory[30,31]. Preliminary phytochemical analysis of *E. scaber* leaves, flower and root and *H. colorata* leaves and stem of crude aqueous, acetone, benzene, chloroform, ethanol and petroleum ether extracts showed the presence of carbohydrates, phenolic compounds, saponins, xanthoprotein, alkaloids, tannins, carboxylic acid, coumarin and flavonoids. It suggests that the plants can be used as anti-microbial activity, anti-oxidant, anti-allergic, anti-inflammatory, antidiabetic, anti-carcinogenic, anti-cancer agents in the future.

E. scaber root ethanol extracts shows highest activity (8/12) against the bacterial pathogens followed by *E. scaber* leaves ethanol extracts (7/12) and *E. scaber* root petroleum ether extracts (6/12). There are some studies on phytochemistry and anti-bacterial activity studies against *E. coli*, *S. aureus*, *E. faecalis*, *P. aeruginosa* and *P. mirabilis* on ethanolic and acetone extracts of *E. scaber* leaves[4,6], but there is no report on against *K. pneumonia*, *S. typhi*, *B. cereus*, *S. marcescens*, *S. pyogenes*, *Acinetobacter* sp. *Enterobacter* sp. In the present study we observed the antibacterial activity against *K. pneumonia*, *S. typhi*, *B. cereus*, *S. marcescens*, *S. pyogenes*, *Acinetobacter* sp. *Enterobacter* sp. But there is no report on *H. colorata* extracts. Thus, the present study shows the presence of antibacterial activity in *H. colorata* extracts for the first time. In the case of *E. scaber*, in addition to

the previous observation, the present study revealed and supplemented the antibacterial activity against the bacterial pathogen *K. pneumonia, S. typhi, B. cereus, S. marsecens, S. pyogenes, Acinetobacter* sp. *Enterobacter* sp.

In the present study, *in vitro* antibacterial efficacy of the *E. scaber* leaves, flower and root and *H. colorata* leaves and stem of crude aqueous, acetone, benzene, chloroform, ethanol and petroleum ether extracts was quantitatively assessed on the basis of zone of inhibition. All the plant parts studied in the present investigation exhibited varying degree of inhibitory effect against the selected bacterial human pathogens. It has been shown that when solvents like ethanol, hexane and methanol are used to extract plants, most of them are able to exhibit inhibitory effect on both gram positive and gram negative bacteria[16]. In the present study also the ethanol, acetone, aqueous, chloroform, benzene and petroleum ether extracts of the selected plants showed zone of inhibition against the isolated human pathogens with varied diameter.

K. pneumoniae is an important cause of human infections and several diseases viz., urinary tract infections, noscomial infections, pneumonia, septicemias and soft tissue infections. The diseases caused by *K. pneumoniae* can result in death of patients who are immunodeficient[32]. In the present study ethanolic extracts of *H. colorata* displayed antibacterial activity against the *K. pneumonia*. It suggested that the plants can be used to treat urinary tract infections, noscomial infections, pneumonia, septicemias and soft tissue infections. *S. pyogenes* causes many important human diseases, ranging from mild superficial skin infections to life-threatening systemic diseases. Infections typically begin in the throat or skin. Examples of mild *S. pyogenes* infections include pharyngitis ("strep throat") and localized skin infection ("impetigo"). Erysipelas and cellulitis are characterized by multiplication and lateral spread of *S. pyogenes* in deep layers of the skin. *S. pyogenes* invasion and multiplication in the fascia can lead to necrotizing fasciitis, a potentially life-threatening condition requiring surgical treatment[33-41]. The ethanolic, chloroform and petroleum ether extracts of *E. scaber* leaves and root demonstrated the antibacterial activity against *S. pyogenes*, it inferred that the selected three plants can be used to treat pharyngitis, impetigo, erysipelas and cellulitis.

The pathogen *S. typhi* is known to cause fever and food borne illness. In the present study the ethanolic extract of roots and leaves of *E. scaber*, leaves and stem extracts of *H. colorata* show the inhibitory activity against the bacteria *S. typhi* and thus the present study confirms the presence of active constituents present in these plants. The result of the present study revealed that the *E. scaber* leaves and root ethanol extracts can be used in the treatment of boils, sores and wounds, since *P. aeruginosa* have been implicated as causative agents of these diseases. Virulent strains of *E. coli* can cause gastroenteritis, urinary tract infections and neonatal meningitis. In rare cases, virulent strains are also responsible for haemolytic uremicsyndrome, peritonitis, mastitis, septicaemia and gram negative pneumonia[42]. In the present study the ethanolic extract of roots of *E. scaber* detected the inhibitory activity against the bacteria *E. coli*. It suggest that the plant can be used to treat urinary tract infections and neonatal meningitis in the future.

B. cereus is responsible for a minority of foodborne illnesses (2%-5%), causing severe nausea, vomiting and diarrhea[43]. In the present study chloroform extracts of *E. scaber* root and H. colorata stem and ethanol extracts of *H. colorata* leaves displayed the inhibition zone against the bacteria *B. cereus*. The present study confirms the folkloric usage of these plants and hint that the selected plants can be used to treat nausea, vomiting and diarrhea. *S. marcescens* can cause infection in several sites, including the urinary tract, respiratory tract, wounds, and the eye, where it may cause conjunctivitis, keratitis, endophthalmitis, and tear ductinfections. It is also a rare cause of endocarditis and osteomyelitis, pneumonia and meningitis[44]. In the present investigation we detected petroleum ether extracts of *E. scaber* and *H. colorata* moderate anti-bacterial activity against the pathogen *S. marcescens*. The results of the present study confirmed the folkloric usage of the plants and previous observations[1,2,5,8]. *E. faecalis* can cause endocarditis, as well as bladder, prostate, and epididymal infections; nervous system infections are less common[45]. The leaves and root ethanolic extracts of *E. scaber* illustrated the antibacterial activity against the bacteria *E. faecalis* and the result of the present study confirms Ahmad observations[3]. Thus the present study confirms the potential value of the two medicinally important plants by the presence of various compounds.

It is hoped that this study would direct to the establishment of some compounds that could be used to invent new and more potent antimicrobial drugs of natural origin. Further work will emphasize the isolation and characterization of active principles responsible for bio-efficacy and bioactivity.

Conflict of interest statement

We declare that we have no conflict of interest.

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